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(54) Title: METHODS AND REAGENTS TO DETECT AND CHARACTERIZE NORWALK AND RELATED VIRUSES

(57) Abstract

Double-stranded cDNA was synthesized from nucleic acid extracted from Norwalk virus purified from stool specimens of volunteers. One clone was isolated from a cDNA library constructed in a pUC-13 vector after amplification of the cDNA. The specificity of this cDNA (pUCNV-953) was shown by hybridization assays. The cDNA reacted with post- (but not pre-) infection stool samples from Norwalk volunteers and with highly purified Norwalk virus, but not with other common enteric viruses such as hepatitis a virus and rotavirus. Finally, the probe detected virus in the same fractions of CsC1 gradients in which viral antigen was detected using a specific Norwalk virus radioimmunoassay, and particles were detected by immune electron microscopy. The availability of a Norwalk-specific cDNA and the first partial genome sequence information allow rapid cloning of the entire genome and of establishment of sensitive diagnostic assays. Such assays can be based on detection of Norwalk virus nucleic acid or Norwalk viral antigen using polyclonal or monoclonal antibodies to proteins expressed from the cDNA or to synthetic peptides made based on the knowledge of the genome sequence. Vaccines made by recombinant DNA technology are now feasible.

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